HEMOCUE®, AN ACCURATE BEDSIDE METHOD OF HEMOGLOBIN MEASUREMENT?

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ABSTRACT. Objective. Evaluate the accuracy of this bedside method to determine hemoglobin (Hb) concentration in general surgery over a wide range of Hb values and to determine potential sources of error. **Methods.** Accuracy of Hb measurement using HemoCue® (AB Leo Diagnostics, Helsinborg, Sweden) was assessed in 140 surgical blood samples using 7 HemoCue® devices in comparison with a CO-Oximeter (IL 482, Instrumentation Laboratory, Lexington, MA). To analyze potential sources of error, packed red cells and fresh frozen plasma were reconstituted to randomized Hb levels of 2–18 g/dL. **Results.** In the surgical blood samples, the Hb concentration determined by the CO-Oximeter (HbCOOX) ranged from 5.1 to 16.7 g/dL and the Hb concentration measured by HemoCue® (HbHC) from 4.7 to 16.0 g/dL. Bias (HbCOOX − HbHC) between HbCOOX and HbHC was 0.6 ± 0.6 g/dL (mean ± SD) or 5.4 ± 5.0% ($p < 0.001$). Also in the reconstituted blood, the bias between HbCOOX and HbHC was significant (0.2 ± 0.3 g/dL or 2.1 ± 3.2%; $p < 0.001$). The microcuvette explained 68% of the variability between HbCOOX and HbHC. HemoCue® thus underestimates the Hb concentration by 2–5% and exhibits a 8–10 times higher variability with only 86.4% of HbHC being within ± 10% of HbCOOX. **Conclusion.** Although the mean bias between HbCOOX and HbHC was relatively low, Hb measurement by HemoCue® exhibited a significant variability. Loading multiple microcuvettes and averaging the results may increase the accuracy of Hb measurement by HemoCue®.

KEY WORDS. Hemoglobin measurement, HemoCue®, hemoglobinometer, CO-Oximeter, spectrophotometer.

INTRODUCTION

Avoiding allogeneic blood transfusions is an important issue in the perioperative care of surgical patients [1, 2]. Low hemoglobin values thus are frequently tolerated. This mandates a fast and reliable method of hemoglobin determination. In many centers, however, blood samples have to be transferred to a laboratory area remote of the operating theater for hemoglobin determination. Delays in analyzing hemoglobin levels might contribute to a prolonged exposure of a patient to a very low hemoglobin level with potential untoward consequences. HemoCue® (AB Leo Diagnostics, Helsinborg, Sweden) represents a bedside hemoglobin measurement device with acceptable accuracy in cardiac surgery [3, 4] as well as in fetal [5, 6], pediatric [7, 8], and outpatient units [9, 10].

The purpose of this study was twofold: First, to evaluate the accuracy of this bedside hemoglobin meas-

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urement technique over a wide range of Hb values in general surgery patients during hemodilution, surgical blood loss and red blood cell transfusion and second, to analyze potential sources of error. Analyzing potential sources of error is important because in most studies in which the overall bias between HemoCue® and the reference method hemoglobin concentrations was considered acceptable, a significant variability was observed between the HemoCue® and the reference hemoglobin concentrations [4, 7–9]. To analyze potential sources of error, heparinized packed red cells and fresh frozen plasma were reconstituted to randomized Hb levels of 2–18 g/dL for specific comparisons between HemoCue® and CO-Oximeter hemoglobin measurements. There were no exclusion criteria for this study.

MATERIALS AND METHODS

With approval of the Ethics Committee, 140 surgical blood samples were simultaneously measured with a CO-Oximeter (IL 482, Instrumentation Laboratory, Lexington, MA) and one of 7 HemoCue® devices (AB Leo Diagnostics, Helsinborg, Sweden) resulting in 20 measurements with each of the HemoCue® devices. The HemoCue® devices were used in random sequence. With each HemoCue® device, all 20 measurements were performed before the next device was tested. The HemoCue® device uses a modified azide methemoglobin reaction [6–8] for hemoglobin quantification. Following red-cell lysis, sodium nitrate converts hemoglobin to methemoglobin, which is then combined with azide. The adsorbance of the azide methemoglobin is then photometrically measured at 565 nm and 880 nm. The CO-Oximeter used in the present study is a micro-computer controlled system which measures the Hb concentration with monochromatic light at four specific wavelengths (535 nm, 585 nm, 595 nm, 627 nm) [11–13]. All hemoglobin measurements were performed immediately after sampling using a single 2-ml syringe of blood that was mixed continuously. All measurements were performed by a single operator.

To analyze potential sources of error, heparinized packed red cells and fresh frozen plasma were reconstituted to randomized Hb levels of 2–18 g/dL (in steps of 2 g/dL). At each hemoglobin level 10 microcuvettes were loaded and the hemoglobin concentration was determined by all 7 HemoCue® devices in randomized order. Simultaneously, the hemoglobin concentration was measured 10 times with the CO-Oximeter. All microcuvettes were measured with all HemoCue® devices resulting in 70 measurements at each Hb level. All microcuvettes were inspected after being loaded to ensure there were no bubbles in the light path [9, 10]. By using new microcuvettes we prevented humidity to interact with the dried-reagent-coated microcuvettes [14]. In both series, the HemoCue® devices used were checked with the device-specific, calibrated mircocuvette before and after use.

Data (mean ± SD) of the surgical blood samples as well as of the reconstituted blood were compared using bias analysis. A potential learning effect was sought in the surgical blood samples by analyzing bias and variability between HemoCue® and CO-Oximeter hemoglobin concentrations in relation to the sequence in which these measurements were performed using analysis of variance (ANOVA) with post-hoc Bonferroni-Dunn correction.

The difference between HemoCue® and CO-Oximeter hemoglobin concentrations of the reconstituted blood was further analyzed by variance components analysis and Wilcoxon's signed rank test. Thereby, a three way analysis of variance with the factors Hb level, HemoCue® device and microcuvette nested in Hb level was performed. Variability of the methods was compared in the reconstituted blood samples using absolute residuals for each Hb level (2 to 18 g/dL).

RESULTS

All HemoCue® devices always measured the hemoglobin concentration of the calibrated microcuvette within ±0.1 g/dL.

In the surgical blood samples, the Hb concentration determined by the CO-Oximeter (HbCOOX) ranged from 5.1 to 16.7 g/dL and the Hb concentration measured by HemoCue® (HbHC) from 4.7 to 16.0 g/dL. Bias (HbCOOX − HbHC) between HbCOOX and HbHC was 0.6 ± 0.6 g/dL (Figure 1A) or 5.4 ± 5.0% (Figure 1B) ($p < 0.001$, each). The difference between the two measurement techniques (HbCOOX − HbHC) exceeded 1.0 g/dL in 25 of 140 samples (17.9%) but never fell below −1 g/dL. In 121 of 140 paired measurements (86.4%), the HbHC was within ±10% of HbCOOX. A significant ($p < 0.001$) learning effect was found such that bias and variability between HemoCue® and CO-Oximeter hemoglobin concentrations were higher in the first 40 measurements compared with the subsequent 100 comparative measurements. However, no further improvement was found in these subsequent 100 measurements (Figure 2).

Also in the reconstituted blood, the bias between HbCOOX and HbHC was significant (0.2 ± 0.3 g/dL (Figure 3A) or 2.1 ± 3.2% (Figure 3B); $p < 0.001$). The difference between the two measurement techniques

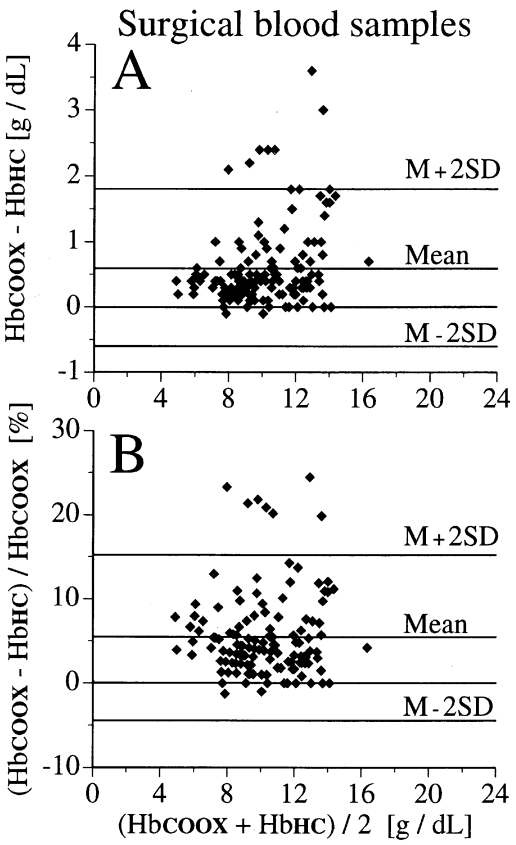

Fig. 1. Bias between Hb concentration measured by CO-Oximeter (HbCOOX) and Hb concentration determined by HemoCue® (HbHC) compared to mean Hb concentration ((HbCOOX + HbHC)/2) in 140 surgical blood samples. (A) absolute difference and (B) percent difference. Indicated are mean bias (Mean), mean + two standard deviations (M + 2SD) and mean − two standard deviations (M − 2SD).

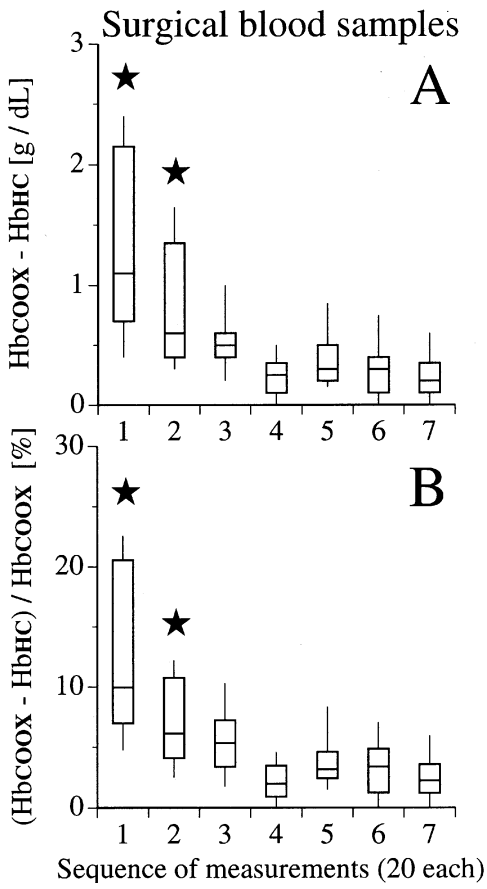

Fig. 2. Box plots of bias between Hb concentration measured by CO-Oximeter (HbCOOX) and Hb concentration determined by HemoCue® (HbHC) in relation to the sequence of measurements (1 through 7; 20 surgical blood samples each). The top, bottom and line through the middle of the box correspond to the 75th, 25th and 50th percentile (median) respectively. The whiskers at the bottom and on top of the box extend to the 10th and 90th percentile. (A) absolute difference and (B) percent difference. The learning effect is documented by a significantly higher bias and variability (★ = $p < 0.05$) in the first 40 measurements compared with the subsequent 100 measurements.

(HbCOOX − HbHC) exceeded 1.0 g/dL in 6 of 630 samples (0.95%) but never fell below −1 g/dL. All HbHC measurements were within ±10% of HbCOOX. Variability of hemoglobin measurement using HemoCue® was 8–10 times higher compared with hemoglobin determination by the CO-Oximeter ($p < 0.001$).

In the reconstituted blood, we also computed variance components for the difference between HbHC and HbCOOX using a three way ANOVA with the factors Hb level, device and microcuvette nested in Hb level. All effects were highly significant ($p < 0.0001$). The microcuvette was found by far the most important factor explaining 68% of the variability between HbCOOX and HbHC followed by Hb level (13%) and HemoCue® device (5%).

DISCUSSION

HemoCue® underestimates the Hb concentration as determined by a CO-Oximeter by 2–5% and exhibits a significantly higher variability. The microcuvette is by far the most important factor explaining the difference between HbCOOX and HbHC. Thus, loading multiple microcuvettes and averaging the results may increase accuracy and reliability of Hb measurement using HemoCue®.

The IL-482 CO-Oximeter used in the present study as the reference method is a micro-computer controlled system which measures the Hb concentration with

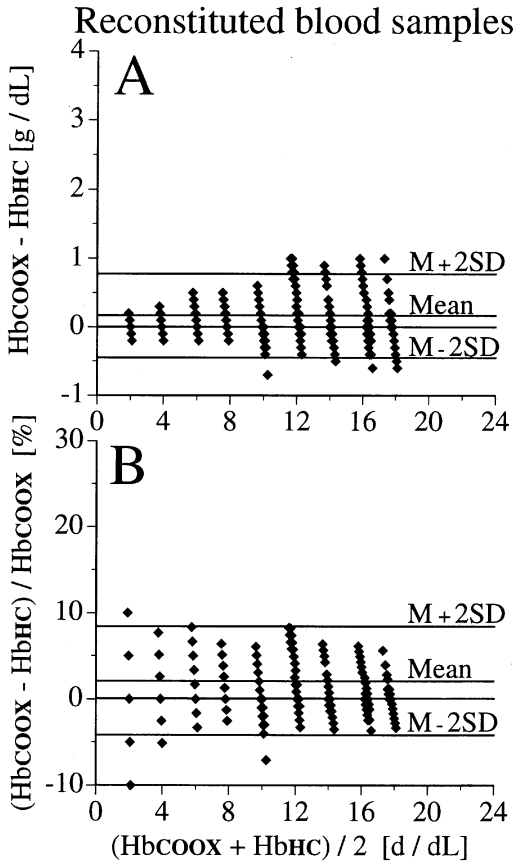

Fig. 3. Bias between Hb concentration measured by CO-Oximeter (HbCOOX) and Hb concentration determined by HemoCue® (HbHC) compared to mean Hb concentration ((HbCOOX + HbHC)/2) in 630 reconstituted blood samples. (A) absolute difference and (B) percent difference. Indicated are mean bias (Mean), mean + two standard deviations (M + 2SD) and mean − two standard deviations (M − 2SD).

monochromatic light at four specific wavelengths (535 nm, 585 nm, 595 nm, 627 nm). Its accuracy is comparable to that of established central hematology laboratory methods [11–13] with a bias between the cyanmethhemoglobin method and the IL-482 CO-Oximeter of 0.1 ± 0.5 g/dL [12] and a nearly ideal linear regression equation of 0.99x + 0.7 (r2 = 0.99) [13]. However, only limited information is available on the variability of the IL-482 CO-Oximeter. In the present study a minimal variability was documented when the reconstituted blood with hemoglobin concentrations ranging from 2 to 18 g/dL was measured 10 times at each hemoglobin concentration with the IL-482 CO-Oximeter and standard deviations of 0.0 g/dL (8 of 9 hemoglobin concentrations) and 0.1 g/dL (1 of 9 hemoglobin concentration) were found. The variability between the HemoCue® and IL-482 CO-Oximeter hemoglobin concentration observed in the present study thus is caused nearly exclusively by the variability of the HemoCue® methodology.

The HemoCue® is a convenient bedside device using a modified azide methemoglobin reaction [6–8] for hemoglobin quantification. Following red-cell lysis, sodium nitrate converts hemoglobin to methemoglobin, which is then combined with azide. The adsorbance of the azide methemoglobin is photometrically measured at 565 nm and 880 nm within 15–45 seconds. The system uses small dried-reagent-coated disposable microcuvettes in which 10 μL of blood is drawn by capillary action. Although handling of the HemoCue® device appears trivial, a distinct learning effect was found in the present study (Figure 2). However, even after the initial learning effect, a 8–10 times greater variability of the HemoCue® methodology remains as compared with the IL-482 CO-Oximeter as documented in the analysis of the reconstituted blood which was performed after the measurements in the surgical blood samples.

Despite avoiding preventable errors in the handling of the HemoCue®, such as air bubbles in the light path [9, 10] or using microcuvettes not stored in a dry environment [14], we found a systematic underestimation of the Hb concentration by HemoCue® of 2.1 ± 3.2% in reconstituted blood and 5.4 ± 5.0% in surgical blood samples. This underestimation is in keeping with previous reports [3–10]. For most clinical applications an average underestimation of the Hb concentration of approximately 5% appears acceptable. However, the variability of the HemoCue® device was 8–10 times higher compared with the CO-Oximeter resulting in a difference of more than 10% in nearly one of six clinical measurements. Should this not be acceptable, loading multiple microcuvettes and averaging the results may decrease this discrepancy since 68% of the difference between HemoCue® and CO-Oximeter readings were found to be due to the microcuvette.

Due to the systematic underestimation, the true Hb concentration is likely to be similar or higher than the Hb concentration displayed by the HemoCue® device. In clinical situations in which a 10% accuracy is acceptable, the HemoCue® device thus may be used as a quick bedside screening method for Hb concentration measurement before sending a blood sample to a central laboratory in order to avoid prolonged exposure of a patient to a very low hemoglobin level with potential untoward consequences.

REFERENCES

1. Rutherford CJ, Kaplan HS. Autologous blood donations – can we bank on it? N Engl J Med 1995; 332: 740–742
2. Spahn DR, Leone BJ, Reves JG, Pasch T. Cardiovascular and coronary physiology of acute isovolemic hemodilution: a review of nonoxygen-carrying and oxygen-carrying solutions. Anesth Analg 1994; 78: 1000–1021
3. McNulty SE, Torjman M, Grodecki W, Marr A, Schieren H. A comparison of four bedside methods of hemoglobin assessment during cardiac surgery. Anesth Analg 1995; 81: 1197–1202
4. Be WK, Kerkkamp HE, Booij LH. Hemocue – a new haemoglobinometer in the clinic. Eur J Anaesthesiol 1991; 8: 55–58
5. Laifer SA, Kuller JA, Hill LM. Rapid assessment of fetal hemoglobin concentration with the HemoCue system. Obstet Gynecol 1990; 76: 723–724
6. Berry SM, Dombrowski MP, Blessed WB, Bichalski JA, Jones TB, Cotton DB. Fetal hemoglobin quantitations using the Hemocue system are rapid and accurate. Obstet Gynecol 1993; 81: 417–420
7. Cohen AR, Seidl-Friedman J. HemoCue system for hemoglobin measurement. Evaluation in anemic and nonanemic children. Am J Clin Pathol 1988; 90: 302–305
8. Chen PP, Short TG, Leung DH, Oh TE. A clinical evaluation of the Hemocue haemoglobinometer using capillary, venous and arterial samples. Anaesth Intensive Care 1992; 20: 497–500
9. Neville RG. Evaluation of portable haemoglobinometer in general practice. Br Med J Clin Res Ed 1987; 294: 1263–1265
10. Von Schenck H, Falkensson M, Lundberg B. Evaluation of "HemoCue," a new device for determining hemoglobin. Clin Chem 1986; 32: 526–529
11. Sedor FA, Holleman CM. Evaluation of the IL 482 CO-Oximeter. Clin Chem 1987; 33: 1942–1943
12. Mahoney JJ, Vreman HJ, Stevenson DK, Van Kessel AL. Measurement of carboxyhemoglobin and total hemoglobin by five specialized spectrophotometers (CO-oximeters) in comparison with reference methods. Clin Chem 1993; 39: 1693–1700
13. Vreman HJ, Mahoney JJ, Van Kessel AL, Stevenson DK. Carboxyhemoglobin as measured by gas chromatography and with the IL 282 and 482 CO-Oximeters. Clin Chem 1988; 34: 2562–2566
14. Henderson MA, Irwin MG. High humidity affects HemoCue microcuvette function (letter). Anaesth Intensive Care 1995; 23: 407.1